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Use of Brine Shrimp, Artemia *spp., in Larval Crustacean Nutrition: A Review*

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ABSTRACT: Because of convenience in production and their suitable biochemical composition, brine shrimp *Artemia* spp. nauplii have been adopted as a standard diet in the commercial larviculture of several crustacean species. The nutritional value of *Artemia*, however, is not constant, but varies both geographically and temporally. During the past decade both the causes of *Artemia* nutritional variability and methods to improve poor-quality *Artemia* have been identified. Enriching *Artemia* spp. with emulsified lipophilic products is a technique that has allowed delivery of extra doses of essential nutrients, for example, highly unsaturated fatty acids (HUFA) and vitamins, to crustacean larvae. The enrichment technique has limitations, however, because the *Artemia* spp. currently available selectively catabolize some of the nutrients such as docosahexaenoic acid and phospholipids. Decapsulated *Artemia* cysts, juveniles, and adult brine shrimp are also used increasingly as suitable diets for different crustacean species.

KEY WORDS: larviculture, shrimp *Penaeus* spp., prawn *Macrobrachium rosenbergii*, n-3 HUFA, vitamin C, vitamin E.

I. INTRODUCTION

Although the brine shrimp *Artemia* sp. has been known to man for centuries, its use as a food for the culture of larval organisms apparently began only in the 1930s to the 1940s. Several investigators found that it made an excellent food for newly hatched fish (Seale, 1933) and shrimp larvae (Hudinaga, 1942). As aquaculture developed in the 1960s and 1970s, the use of *Artemia*, although not a natural diet, also became more widespread, due both to convenience and to the nutritional value of *Artemia* for larval organisms. The fact that dormant cysts of *Artemia* can be stored for long periods in cans, then used as an off-the-shelf food requiring only 24 h of incubation, makes them the most convenient and least labor-intensive live food available for the culture of several crustacean species, for example, various species of shrimp, lobster, and crab (Léger et al., 1986). In the late 1970s, and again recently, cyst supplies have not been reliable, resulting in fluctuating prices and unforeseen shortages. Furthermore, the nutritional value of *Artemia*, especially for marine fish

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and crustaceans, is not constant, but varies geographically and temporally (Léger et al., 1986).

During the last decade some of the causes of *Artemia* nutritional variability and methods to improve poor-quality *Artemia* have been identified. This article covers historical and present usage of *Artemia* in crustacean larviculture, with particular emphasis on the nutritional aspects of this important food. More in depth reviews of some of the subjects covered here can be found in Sorgeloos (1980), Watanabe et al. (1983), Léger et al. (1986), and Bengtson et al. (1991).

II. CYST SUPPLY AND DEMAND

The initial sources of commercial cysts were a coastal saltwork in the San Francisco Bay (SFB), California (USA), in the 1950s and later from the inland Great Salt Lake (GSL) in Utah (USA). Cyst prices increased considerably in the mid-1970s as the combined result of increased demands from the emerging hatchery industry, decreased harvests from the Great Salt Lake, and possibly simulated shortages by certain commercial companies. Based on research performed at the State University of Gent in Belgium, the idea was launched at the 1976 FAO Technical Conference on Aquaculture in Kyoto (Japan) that the cyst shortage was an artificial and temporary problem that could be overcome by the exploration for and development of new *Artemia* resources and by the application of improved methods for processing and use of the cysts (Sorgeloos, 1979).

By 1980 the situation was rectified with several new commercial products from natural (e.g., Australia, France, PR China) and man-managed (e.g., Brazil, Thailand) *Artemia* production sites. Hatching rates and nutritional quality of cysts were variable, but during the 1980s new methods for collecting and processing the cysts and for evaluating and manipulating the nutritional composition of the nauplii were adopted. In the meantime cyst consumption had increased exponentially as a result of the booming shrimp and fish hatchery industry. Presently, some 5000 fish and shrimp hatcheries require over 1000 metric tons of cysts annually (Van Stappen and Sorgeloos, 1993). Strong competition in the marketplace especially with cyst products that could be harvested very efficiently and cost-effectively from the Great Salt Lake resulted in a new vulnerable situation of dependence on one resource. The warnings of Bengtson et al. (1991) were neglected and the poor yield from the Great Salt Lake during the 1993 to 1995 cyst harvesting seasons resulted in a severe cyst shortage in 1995 (Sorgeloos and Van Stappen, 1995). As cyst prices increase it will become attractive again to explore and develop new *Artemia* resources or to switch to more cost-effective formulated feeds (Sorgeloos and Léger, 1992).

III. PRODUCTION AND USE OF FRESHLY HATCHED NAUPLII

Although using *Artemia* cysts appears to be simple, several factors are critical for hatching the large quantities needed in larval crustacean production. These include cyst disinfection or decapsulation prior to incubation and hatching under the

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following optimal conditions: constant temperature of 25 to 28°C, 15 to 35 ppt salinity, minimum pH of 8.0, near saturated oxygen levels, maximum cyst densities of 2 g/l, and strong illumination of 2000 lux (Sorgeloos et al., 1986). All these factors will affect the hatching rate and maximum output and hence the production cost of the harvested *Artemia* nauplii. Attention should be paid to select *Artemia* cyst lots with good hatching synchrony (less than 7 h between hatching of first and last nauplii) and high hatching efficiency (more than 200,000 nauplii per gram product), as considerable variation has been demonstrated cysts of various origin, and even among batches from the same strain.

After hatching, and prior to feeding them to the larvae, *Artemia* nauplii should be separated from the hatching wastes. After switching off the aeration in the hatching tank, cyst shells will float and nauplii will concentrate at the tank's bottom. They should be siphoned off within 5 to 10 min and thoroughly rinsed with seawater or freshwater, using submerged filters (Sorgeloos and Léger, 1992) to prevent physical damage to the nauplii.

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IV. SIZE AND ENERGY CONTENT

In their first stage of development, *Artemia* nauplii do not feed but consume their own energy reserves (Benijts et al., 1976). At the high water temperatures that are applied during cyst incubation, freshly hatched *Artemia* naupli develop into the second larval stage within 6 to 8 h. It is important to feed first-instar naupli rather than starved second-instar metanauplii that are transparent and less visible. Instar II metanauplii are about 50% larger in length and swim faster than first instars. As a result they are less acceptable as prey. Furthermore, they contain lower amounts of free amino acids so they are less digestible and their lower individual dry weights (1.63 μg vs. 2.15 μg in the SFB strain) and energy content (0.0366 Joules vs. 0.0500 Joules in the same strain) reduce the energy uptake by the predator per unit of hunting effort (Léger et al., 1986). All this will be reflected in reduced larval growth in the face of increased *Artemia* cyst consumption (20 to 30% more cysts are needed to feed the same weight of starved metanauplii to the predator).

Storing freshly hatched nauplii at temperatures near 4°C, in densities of up to eight million nauplii per liter for up to 24 h (Léger et al., 1983) will greatly reduce their metabolic rate, that is, only a 2.5% drop in individual dry weight vs. 30% at 25°C, and preclude molting to the second instar stage. The 24–h cold storage economizes the *Artemia* cyst hatching effort (e.g., fewer tanks, larger volumes, a maximum of one hatching and harvest per day) and allows not only a constant supply of a high-quality product but also the possibility of more frequent food distributions. This is beneficial for shrimp larvae because food retention time in larviculture tanks can be reduced and hence the growth of *Artemia* in the culture tank minimized. With poor hunters, such as the larvae of the tiger shrimp *Penaeus monodon*, feeding cold-stored and thus less active *Artemia*, results in much more efficient food uptake. Although the same is true when using frozen *Artemia* nauplii (Mock et al., 1980), this procedure might result in nutrient losses as the freezing process physically damages the *Artemia* nauplii. Because penaeid shrimp larvae apparently benefit from a zooplankton diet in the zoea stages (Mock et al., 1980; Wilkenfeld et al., 1984) *Artemia* is sometimes introduced to the zoea-III stage in the

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form of decapsulated cysts or as freshly hatched nauplii that have been killed by a very short heat-shock treatment (i.e., dipping of a net with the *Artemia* nauplii in 80°C water). As the size of *Artemia* cysts and consequently their nauplii can vary significantly (Vanhaecke and Sorgeloos, 1980), it is critical to choose a strain with small cysts (diameter < 230 μm) for use in start-feeding crustacean larvae.

V. NUTRITIONAL QUALITY — FATTY ACID ENRICHMENT

In the late 1960s and early 1970s several authors reported problems in larviculture success with shrimp, prawn, lobster, and crab species when using *Artemia* sources other than SFB *Artemia* (for reviews see Sorgeloos, 1980 and Léger et al., 1986). High doses of toxic compounds (e.g., chlorinated hydrocarbons and heavy metals), were initially suspected to be the cause of the poor nutritional value of *Artemia* from GSL and the People's Republic of China. A comparative study with eight strains of *Artemia* spp. using crab and mysid shrimp as predator test species confirmed the nutritional variation among *Artemia* sources (Johns et al., 1980, 1981; Léger and Sorgeloos, 1984). Léger et al. (1985a) documented the nutritional variability in 11 batches of San Francisco Bay *Artemia* nauplii for the mysid shrimp *Mysidopsis bahia*. Similar to findings by Watanabe et al. (1978) and Kanazawa et al. (1979) in marine fish, Léger et al. (1985b, 1987a) concluded that the main factor affecting the nutritional value of *Artemia* for marine shrimp larvae was the content of the highly unsaturated fatty acid (HUFA) eicosapentaenoic acid 20:5n-3 (EPA).

Taking advantage of the primitive feeding characteristics of *Artemia* nauplii, it is possible to manipulate the nutritional value of HUFA-deficient *Artemia*, for example, the GSL strain. Because brine shrimp nauplii that have molted into the second instar stage (i.e., about 8 h following hatching) are nonselective particle feeders, simple methods have been developed to incorporate different kinds of products into the *Artemia* prior to feeding to predator larvae. This method of "bioencapsulation", also called *Artemia* enrichment or boosting, is widely applied in marine fish and crustacean hatcheries for enhancing the nutritional value of *Artemia* with essential fatty acids.

British, Japanese, and Belgian researchers developed enrichment products and procedures using selected microalgae and/or microencapsulated products, yeast and/or emulsified preparations, self-emulsifying concentrates, and/or microparticulate products (Léger et al., 1986). The highest enrichment levels are obtained from emulsified concentrates: freshly hatched nauplii are transferred to the enrichment tank at a density of 100 to 300 nauplii/ml (for enrichment periods >24 h or <24 h, respectively). The enrichment medium consists of hypochlorite-disinfected and neutralized seawater maintained at 25°C. The enrichment emulsion is added in consecutive doses of 0.3 g/l every 12 h. Strong aeration using airstones or pure oxygen is required to maintain dissolved oxygen levels above 4 ppm. Enriched nauplii are harvested after 24 or 48 h, thoroughly rinsed and stored at temperatures below 10°C in order to assure that HUFA are not metabolized during storage. Enrichment levels of 50 to 60 mg/g DW n-3 HUFA are obtained after 24 h enrichment with the emulsified concentrates. Nauplii should be transferred or exposed to the

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enrichment medium as soon as possible before first feeding so they begin feeding immediately after the opening of the alimentary tract (instar II stage). As a result, the increase of nauplius size during enrichment can be minimized, that is, after 24 h enrichment GSL *Artemia* nauplii will reach about 660 μm, and after 48 h enrichment about 790 μm.

Feeding n-3 HUFA-enriched *Artemia* nauplii results in increased larval survival and growth in several *Penaeus* spp. and *Macrobrachium rosenbergii* (Bengtson et al., 1991). In addition, Léger and Sorgeloos (1992) showed that in penaeid shrimp the effect of diet composition, especially with respect to n-3 HUFA content, might only become evident in later stages: the negative effects on survival and growth when feeding n-3 HUFA-poor *Artemia* were aggravated when the animals had previously been fed a n-3 HUFA-poor diet as opposed to an n-3 HUFA-rich food during the zoeal stages. When postlarvae that had been fed with enriched GSL *Artemia* were subsequently fed an artificial diet, better food acceptance, growth, and survival were recorded than in postlarvae previously fed n-3 HUFA-poor *Artemia*.

Another illustration of this delayed dietary effect is the resistance to salinity stress in PL-10 stages of a batch of *Penaeus monodon* larvae fed on three different larval diets that varied in n-3 HUFA levels (Tackaert et al., 1992). Differences in survival among the three diets were not significant at PL-10; however, differences in PL quality, expressed as their ability to survive the applied salinity stress, were pronounced, that is, 90% survival after 1 h exposure to a salinity shock at 7 ppt vs. less than 25% survival in the low-HUFA dietary regimes.

Although the cited studies provided convincing evidence of the importance of the n-3 HUFA in *Artemia* when used as food for shrimp larvae, quantitative dietary requirements as well as the relative importance of selected HUFA (e.g., docosahexaenoic acid, 22:6n-3, DHA) remained to be explored. Rees et al. (1994) fed *Penaeus monodon* postlarvae (PL-5 to PL-15) five diets consisting of *Artemia* nauplii enriched with different n-3 HUFA levels. Although the postlarvae grew well on an *Artemia* diet with low HUFA content, their survival was poor and the PL-10s had limited ability to endure osmotic stress. Feeding *Artemia* enriched with medium levels of 12.5 mg/g DW n-3 HUFA (DHA/EPA ratio of 0.4 in the *Artemia*) considerably enhanced survival of PL-15s (from 20 to 60%) and the resistance of PL-10s to osmotic stress (drop in cumulative mortality index from 90 in the control to less than 30 in the HUFA-enriched series). This was confirmed by Kontara et al. (1995a), who detected significant differences in production characteristics of *P. monodon* (85.8 vs. 61.8% survival, 93.3 vs. 26.7% stress survival at PL-15) when HUFA-fortified *Artemia* (39.2 mg/g DW) compared with low-HUFA *Artemia* (3.5 mg/g DW) were fed from PL-4 onward. These results confirmed the earlier suggestion of Kanazawa (D'Abramo, 1991) that 1% n-3 HUFA in the diet is a minimal value for postlarval penaeids. Rees et al. (1994) found that very high dietary levels of 31 mg/g DW n-3 HUFA (DHA/EPA ratio of 0.5) did not have any growth-promoting effect or improved postlarval stress-resistance, suggesting that an excessive supply of n-3 HUFA may not be beneficial to the shrimp.

Only a few authors have studied n-3 HUFA requirements in *Macrobrachium* sp. Sheen and D'Abramo (1990) demonstrated significantly better growth in juvenile prawn with a HUFA-supplemented experimental diet. Devresse et al. (1990) reported significant increases in both survival and growth, more precocious and more synchronous metamorphosis as well as a higher stress resistance in *M. rosenbergii*

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larvae fed n-3 HUFA-fortified vs. control *Artemia* nauplii. Using *Artemia* enriched with different n-3 HUFA emulsions, made available in the framework of an international intercalibration study (ICES, 1988), Romdhane et al. (1995) showed that the minimum requirement for larval *M. rosenbergii* is about 35 mg/g DW total n-3 HUFA in the *Artemia* diet. Recently, however, de Caluwe et al. (1995) demonstrated that these n-3 HUFA effects in larval *Macrobrachium* are also function of the broodstock diet: larvae obtained from females fed a HUFA-fortified diet performed equally well on nonenriched as on enriched *Artemia*.

Recent studies with various species of marine fish have revealed that high dietary levels of total HUFA can have a negative effect, and that DHA is more important than EPA for various physiological functions, including survival, growth, and pigmentation success (Kanazawa, 1993; Koven et al., 1993; Watanabe and Kiron, 1994; Sorgeloos et al., 1995).

In order to verify this hypothesis for different species of shrimp, emulsions were prepared with DHA/EPA ratios varying from 0.6 to 4.0 while maintaining constant HUFA levels (ICES, 1991). Culture tests using enriched *Brachionus* and *Artemia* fed to larval (Z-1 to PL-11) *P. vannamei* (Wouters et al., 1997) and postlarval (PL-5 to PL-25) *P. monodon* (Kontara et al., 1995a) revealed no significant differences in function of the various DHA/EPA ratios for survival, growth and metamorphosis rate. These results indicate that there is no specific requirement for DHA over EPA in larval shrimp. This was confirmed for postlarval *P. vannamei* (PL-10 to PL-35) using semipurified diets that only varied in the DHA/EPA ratio of the lipid fraction (range tested 0.6 to 3.6; 25% n-3 HUFA of total HUFA; Naessens et al., 1995a).

It appears from the former studies and from Dhert et al. (1993a) and Triantaphyllidis et al. (1995) that *Artemia* (at least the most commonly used species *A. franciscana*) is not a suitable experimental live food to study quantitative lipid requirements in larval fish and shellfish. Contrary to other live feeds, such as rotifers, the enrichment of *Artemia franciscana* with DHA is difficult because of the inherent catabolism of this fatty acid after enrichment. DHA/EPA ratios obtained immediately after enrichment with DHA-rich emulsions are considerably lower in *Artemia* than in rotifers. The capability of some Chinese *Artemia* strains to reach high DHA levels during enrichment (Dhert et al., 1993a) and to maintain them during subsequent starvation (Evjemo et al., 1997) offers new perspectives for providing higher dietary DHA levels and DHA/EPA ratios to fish and crustacean larvae.

It would be interesting to use similar *Artemia* treatments with high HUFA and high DHA/EPA ratio to verify larval fatty acid requirements in crustaceans that are going through very pronounced metamorphosis (e.g., crab and spiny lobster species) for which very low survival rates and/or high incidence of abnormal developments have been reported (Kittaka, 1994; Liong, 1995).

VI. PHOSPHOLIPIDS

Although phospholipid requirements are well documented in juvenile stages for various crustacean species only limited information is available on the role of phospholipids in start-feeding stages (reviewed by Coutteau et al., 1997). The few studies on larval requirements of crustacean species used artificial diets (Teshima et al., 1982, Kanazawa et al., 1985, Camara et al., 1997). As shown by Tackaert et al.

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(1991) *Artemia* appears not to be a suitable test diet to study phospholipid requirements, that is, dietary enrichment with phosphatidylcholine (PC) did not enhance the PC content in *Artemia*. Rainuzzo et al. (1994) found similar lipid composition in *Artemia* enriched with an emulsion based on either ethyl esters or halibut roe, containing, respectively, 72.6% neutral lipids (mainly ethyl esters) and 71.2% polar lipids (mainly PC and phosphatidylethanolamine, PE). Still, limited shifts of lipid classes, for example, PC/PE ratio (Rainuzzo et al., 1994), due to enrichment of *Artemia* are poorly documented and their significance in terms of nutritional value unknown.

VII. ***VITAMIN C***

Vitamin C, more specifically ascorbic acid (AA), is generally considered to be an essential dietary component for the various stages of aquaculture organisms. Several biological (e.g., skeletal development, growth, survival) as well as physiological functions (e.g., resistance to toxicants and stress, immunoactivity) are enhanced in larvae from supplemental dietary ascorbate (Dabrowski, 1992; Merchie et al., 1997b). Ascorbic acid 2–sulfate (AAS), a stable derivative of AA, was discovered in dormant cysts of *Artemia* by Mead and Finamore (1969). Cysts of various batches differed considerably in AAS content: 160 to 517 μg/g DW, expressed as AA (Dabrowski, 1991; Merchie et al., 1995a). The amount of AA, liberated in freshly hatched nauplii reflects the AAS reserve present in the cysts and provides evidence for the conversion of AAS to free AA during completion of embryonic development into nauplii (Golub and Finamore, 1972; Dabrowski, 1991; Nelis et al., 1994).

The variation in AAS concentration observed in *Artemia* cysts may reflect adult nutrition during egg production as was demonstrated for HUFA content (Lavens et al., 1989), and this may explain the differences among batches of the same strain (Merchie et al., 1995a). Differences among geographical populations and *Artemia* species and broods from different years might significantly influence the AAS content in the cyst material, thus the AA levels in freshly hatched nauplii and consequently, their nutritional value for larval fish and crustaceans.

Tests have been conducted to incorporate extra AA into *Artemia* nauplii in a stable and bioavailable form. Applying a standard enrichment procedure (Léger et al., 1987b) and experimental self-emulsifying concentrates containing 10 to 20% ascorbyl palmitate (AP), levels up to 2.5 mg free AA/g DW can be incorporated into brine shrimp nauplii within 24 h (Merchie et al., 1995a). These concentrations did not drop when the 24–h enriched nauplii were stored for another 24 h in seawater at 28 or 4°C.

The effect of vitamin C enrichment in *Artemia* nauplii on larviculture outputs has been verified for *Macrobrachium rosenbergii* (Merchie et al., 1995b). A control (550 μg AA/g DW) was compared with two different AA-enrichment levels in *Artemia* (1300 and 2750 μg AA/g DW). Under standard culture conditions, no differences on growth or survival were observed demonstrating that the nutritional requirements are below 550 μg AA/g DW (the normal level occurring in freshly hatched *Artemia)*. However, significantly positive effects on the physiological condition of the postlarvae, measured by a salinity stress test, could be demonstrated when vitamin C-boosted live food was administered.

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Because the AA levels in predator larvae are linked with the enrichment levels in the live prey, it may be assumed that a stress resistance was increased by feeding vitamin C-enriched *Artemia*. Under suboptimal conditions supplementation with high vitamin C levels might also enhance production. These results support the hypothesis that stress creates increased ascorbate requirements for larval fish and crustaceans, and that in this respect body vitamin C concentration may reflect the survival potential more accurately than variation in growth rate (Dabrowski, 1992). Moreover, at day 28, a significant drop in AA concentration was detected in the postlarvae compared with the levels found in the larvae. This may reflect an extra need for vitamin C during metamorphosis, a stressful period as the larvae undergo major morphological and physiological changes.

Recent culture tests with vitamin C boosted microbound diets for *P. vannamei* confirmed the hypothesis that dietary vitamin C improved stress resistance in postlarval crustaceans (Kontara et al., 1995b; Merchie et al., 1996a).

VIII. OTHER NUTRIENTS

High levels of α-tocopherol can be bioaccumulated and maintained in *Artemia* nauplii (Lavens and Nelis, personal communication) making this live food delivery system useful for studying dietary requirements and antioxydative effects of vitamin E in larval crustacean nutrition research. The effectiveness of *Artemia* nauplii as a dietary carrier system could be tested for various other nutritional components, such as, liposoluble products administered via an emulsion, water-soluble compounds via liposomes (Hontoria et al., 1994), and/or microcapsule delivery (Sakamoto et al., 1982). However, for each nutrient the usefulness of the *Artemia* bioencapsulation method remains to be verified by chemical analysis. For example, do the *Artemia* bioaccumulate in their digestive tracts and/or assimilate in their tissues or do they selectively catabolize the test-nutrient?

IX. OTHER FORMS OF ARTEMIA USED IN CRUSTACEAN NUTRITION

Aside from the most common regime of feeding freshly hatched and/or 24–h enriched nauplii, the use of dry decapsulated cysts, juveniles, and adult biomass is practiced with various crustacean species (Léger et al., 1986; Bengtson et al., 1991; Stael et al., 1995). Decapsulated cysts (also called de-shelled or shell-free cysts) can be used in start-feeding crab, shrimp, and prawn larvae; however, the rapid settling of the cysts in seawater can make them unavailable for planktonic larvae unless they can be kept in suspension by using conical-shaped culture tanks and strong aeration. Best results are obtained when feeding decapsulated cysts to postlarval shrimp and prawn as a partial or complete substitute for live nauplii (Stael et al., 1995). The major advantage here might be, aside from being a directly available off-the-shelf product, that cysts with poor hatching quality can still be used as a food source.

Dhert et al. (1993b) developed a simple culture system for juvenile and adult *Artemia* as food for postlarval *Penaeus monodon*. The growth performance of

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shrimp reared from PL-4 to PL-25 on juvenile *Artemia* live prey is identical to the growth obtained when feeding newly hatched *Artemia*. Furthermore, the PL-25 reared with juvenile brine shrimp display significantly better resistance in salinity stress tests. The stress sensitivity index dropped from 138 with freshly hatched nauplii to 36 when feeding juvenile *Artemia*. Besides nutritional and energetic advantages, the use of *Artemia* biomass for feeding postlarval shrimp also results in improved economics as expenses for cysts and weaning diets can be reduced.

The high food value in nursery culturing of shrimp, prawn, and lobster of adult *Artemia* harvested from ponds or from intensive indoor culture systems is well documented. The most spectacular example was the use of thousands of tons of fresh brine shrimp biomass harvested from coastal and inland saltworks as a supplementary natural food in the pond culture of *Penaeus chinensis* in the Bohai Bay, People's Republic of China (Tackaert and Sorgeloos, 1991).

Although the fresh-live form has the highest nutritive value, harvested *Artemia* can also be frozen, freeze-dried, or acid-preserved (Abelin et al., 1991; Naessens et al., 1995b) for later use, or made into flakes or other forms of formulated feed. *Artemia* biomass is apparently a good food for the maturation of several species of penaeid shrimp. Recent culture tests in Ecuador and the USA have shown that polychaetes, which have been identified as a critical fresh-food component in the maturation diet of *Penaeus vannamei* (Bray and Lawrence, 1992) can be successfully replaced by frozen *Artemia* biomass (Naessens et al., 1997).

X. CONCLUSION

Empirical applications with the brine shrimp *Artemia* have resulted in quick and successful developments in the commercial hatchery rearing of several crustacean species. The availability of nutritionally different sources of brine shrimp cysts and the use of simple bioencapsulation techniques with the nauplii have in the past, and can in the future, contribute significantly to a better understanding of the nutritional requirements of larval crustaceans. This knowledge will allow further improvement of the formulation and manufacturing of artificial diets, which eventually should completely replace *Artemia* and other live feeds and lead to more reliable and cost-effective hatchery operations.

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